Solubility and Salting Behavior of Several β-Adrenergic Blocking Agents in Liquid and Supercritical Carbon Dioxide

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The phase behavior of five β-adrenergic blocking agents (acebutolol, atenolol, nadolol, pindolol, and propranolol) were explored in liquid and supercritical carbon dioxide at (298, 308, and 318) K and at pressures between (80 and 275) bar. The solubility of the solids in carbon dioxide was experimentally determined by observing the cloud point using a variable-volume stirred vessel with visual access. Although these compounds are very similar structurally, each containing a secondary amine, alcohol, aromatic ring, and ether group, only pindolol was soluble in carbon dioxide under the conditions explored. The other four β blockers were observed to form yellow-brown salt complexes with carbon dioxide and did not show any appreciable solubility. It is believed that pindolol's lower basicity (having a K_b an order of magnitude lower than the other four compounds) prevented it from reacting with carbon dioxide, a Lewis acid, and salting out as the other four β blockers did.

Introduction

β-Adrenergic blocking agents, or β blockers, inhibit the effects of adrenaline on the body's β receptors. β blockers are used to treat high blood pressure, congestive heart failure, arrhythmias, and angina and are sometimes used in cardiac patients to prevent future heart attacks.[1−4] Cardioselective β blockers predominantly block the β-1 receptors in cardiac tissue. By blocking the β-1 receptors, the nerve impulses that travel through the heart are slowed, and the heart does not work as hard. Nonselective agents block both β-1 receptors and β-2 receptors (primarily located in tissues other than cardiac). Acebutolol and atenolol are cardioselective, whereas nadolol, pindolol, and propranolol are nonselective.[1]

Typical marketed chiral β blockers are racemic mixtures consisting of equal molar mixtures of the (+) and (−) enantiomers.[5] The (−) enantiomers have been shown to have the desired pharmacological activity,[6] whereas the (+) enantiomers tend to increase the toxicity and adverse side effects of the drug because they undergo different metabolic pathways than their more active isomers.[5,7] It would be beneficial to develop separation techniques to isolate the active enatiomer for use in pharmaceutical products.

Compressed carbon dioxide has been shown to be a suitable solvent for use in supercritical fluid chromatography techniques for the separation and purification of many isomeric and enantiomeric pharmaceutical products as highlighted in the review article by Foster et al.[8] Carbon dioxide has even been used recently with methanol as a cosolvent to separate the racemic β blocker propranolol-HCl using supercritical fluid chromatography with UV and polarimetric detection.[9] However, before we can attempt to develop techniques to isolate chiral β blockers, the solubilities of the compounds in liquid and supercritical carbon dioxide must be explored. It is often difficult to predict whether compounds will dissolve in carbon dioxide and if they do, to what extent.

We chose to explore the phase behavior of five common β blockers—acebutolol, atenolol, nadolol, pindolol, and propranolol—in liquid and supercritical carbon dioxide under typical pressure and temperature operating conditions for chromatography. All of these compounds are chiral and of the 3-(aryloxy)-1-(alkylamino)-2-propanol type. This study will provide the data required to design a supercritical fluid chromatography separation procedure properly. All of the β blockers were used in their free-base forms because hydrochloride salts have been shown not to be soluble in pure carbon dioxide.[9,10] It is essential to explore these compounds in carbon dioxide because they all contain a secondary amine group and some of the compounds have other amine groups as well (Figure 1). Francis[11] and Dandge et al.[12] both found that the weakly acidic carbon dioxide can have an affinity for several basic amines, causing salt formation that drastically lowers the solubility or even makes the compounds insoluble in carbon dioxide. Therefore, the salting behavior of the β blockers will need to be explored along with their solubilities as a function of temperature and pressure.

Experimental Section

Materials. Research grade 5 carbon dioxide with a purity of 99.999% was supplied by BOC Gases. Atenolol (CAS 29122-68-7, 4-[2-hydroxy-3-[(1-methylethyl)amino]-propoxy] benzeneacetamide, >98% purity), nadolol (CAS 42200-33-9, 1-(*tert*-butylamino)-3-[(5, 6, 7, 8 -tetrahydro-*cis*-6, 7-dihydroxy-1-naphthyl)oxy]-2-propanol, >98% purity), and pindolol (CAS 13523-86-9, 1-(1H-indol-4-yloxy)-3-(isopropylamino)-2-propanol, 1-(1H−Indol-4-yloxy)-3-[(1-methylethyl)amino]-2-propanol, >98% purity) were supplied by Sigma-Aldrich and used as received as an equal molar racemic mixture of each. Acebutolol hydrochloride (CAS 34381-68-5, *N*-(3-acetyl-4-[2-hydroxy-3-(isopropylamino)propoxy] phenyl)butanamide hydrochloride, >99% purity) and

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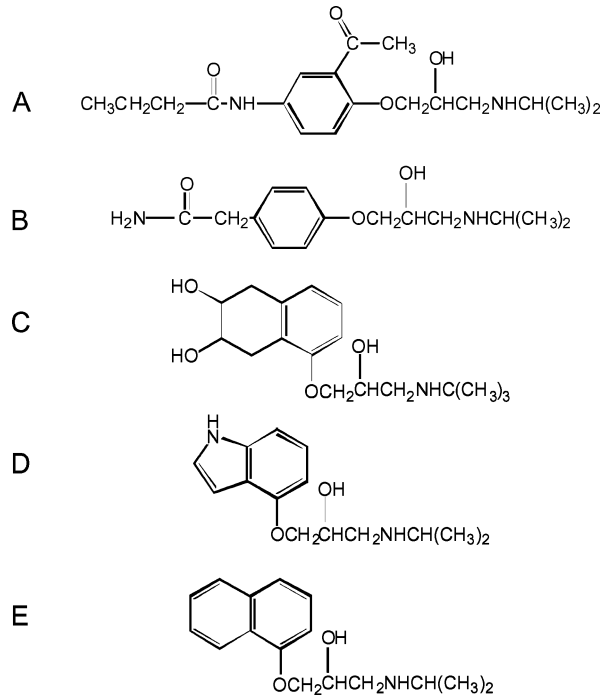

Figure 1. Structures of (A) acebutolol ($C_{18}H_{28}N_2O_4$), (B) atenolol ($C_{14}H_{22}N_2O_3$), (C) nadolol ($C_{17}H_{27}NO_4$), (D) pindolol ($C_{14}H_{20}N_2O_2$), (E) and propranolol ($C_{16}H_{21}NO_2$).

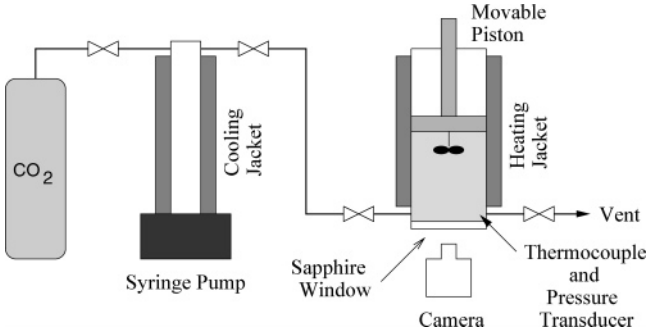

Figure 2. Schematic of the phase equilibrium apparatus.

propranolol hydrochloride (CAS 4199-10-4, 1-(isopropylamino)-3-(1-naphthoxy)-2-propanol hydrochloride, >99% purity), also equal molar racemic mixtures, were obtained from Sigma-Aldrich. All purities are given as weight percent. The hydrochloride was used to make the free base using similar procedures developed by Neau et al.[5] To obtain the free base, we dissolved approximately 0.25 g of the hydrochloride in 5 mL of deionionzed ultrafiltered water to which 1 mL of 5 M NaOH was added, causing the free base to form and drop out of solution, which was extracted with 20 mL of methylene chloride. The solution was placed in the hood until the methylene chloride evaporated. The captured solid was dried in an oven at 45 °C for about an hour. All free-base β blockers were white powders.

Experimental Procedures. A dynamic system manufactured by Thar Design Technologies (PEA-30ML Phase Equilibrium Analyzer) was used to observe the cloud points and salting behavior of the β blockers in carbon dioxide. Details of the experimental apparatus (Figure 2) are presented in our previous paper along with the expected errors in the measurements.[10] In that paper, we also verified our solubility measurement procedures by matching the solubility of ketoprofen in carbon dioxide to that found in the literature.[10,13,14]

Table 1. Mole Fraction Solubility (y) of Pindolol in Liquid and Supercritical Carbon Dioxide

T = 298 K			T = 308 K			T = 318 K		
P bar	ρ g·cm^{-3}	y (10^4)	P bar	ρ g·cm^{-3}	y (10^4)	P bar	ρ g·cm^{-3}	y (10^4)
80	0.778	0.660	80	0.436	0.308	80	0.242	0.337
90	0.801	0.762	90	0.665	0.315	90	0.34	0.270
100	0.819	0.842	100	0.715	0.380	100	0.503	0.282
125	0.852	1.03	125	0.778	0.702	125	0.679	0.695
150	0.877	1.33	150	0.816	0.938	150	0.743	0.875
175	0.898	1.44	175	0.844	1.28	175	0.784	1.08
200	0.915	1.58	200	0.866	1.41	200	0.814	1.21
225	0.93	1.89	225	0.885	1.76	225	0.838	1.55
250	0.944	2.10	250	0.902	2.00	250	0.858	1.78
275	0.956	2.24	275	0.916	2.11	275	0.875	1.92

To observe salting behavior, we placed a small sample (0.0010 g) of a β blocker in the bottom of the vessel (fixed at 6 mL), which was then sealed and flushed with low-pressure carbon dioxide while the vessel was preheated to the desired temperature. Once at the desired temperature, stirring was initiated, and the vessel was quickly pressurized to the desired set point. Salting was observed when the white solids clumped together and changed color to a brownish yellow.

To measure the solubility, we loaded and pressured the vessel as in the salting experiments. The volume of the vessel was then slowly increased by raising the piston while keeping the temperature and pressure constant. Once all of the solid particles were visually observed to dissolve, the vessel was isolated from the pump, and its volume slowly increased until material was seen to drop out of solution (the cloud point). At this point, the temperature, pressure, and volume of the system were recorded. With the known amount of drug placed in the vessel and the density of pure carbon dioxide (calculated using the Thermodynamic and Transport Properties of Pure Fluids: NIST Standard Database 12 Version 5.0[15] with the measured temperature and pressure), the solubility of the drug could be calculated. Because the mole fraction solubilities were found to be on the order of 10^{-4} to 10^{-5}, using properties of pure carbon dioxide was justified. After each experiment, the vessel was cleaned with ethanol and dried.

Results and Discussion

Pindolol was the only one of the five compounds tested that did not react with carbon dioxide to form a salt. The solubility mole fraction of pindolol in carbon dioxide at (298, 308, and 318) K between (80 and 275) bar is presented in Table 1 and Figure 3. As the pressure is increased, the solubility of pindolol increases, as expected. It appears from examining Figure 3 that as temperature increases the solubility of pindolol decreases. However, the density of carbon dioxide and hence its solvating power significantly decreases with increasing temperature. Therefore, it is more appropriate to examine the solubility of pindolol as a function of density as opposed to pressure. Figure 4 clearly shows that at constant density an increase in temperature slightly increases the solubility of the drug in carbon dioxide. This increase in solubility is expected[10,16] as the sublimation pressure of the drug, driving it into the fluid phase, increases with an increase in temperature. There does appear to be a large jump in solubility above 0.65 g·cm^{-3} at all three temperatures. However, we believe this to be caused by the error in weighing the small masses of drug required to obtain the low mole fraction readings rather than any physical interactions between the drug and carbon dioxide at the higher densities. The amount weighed

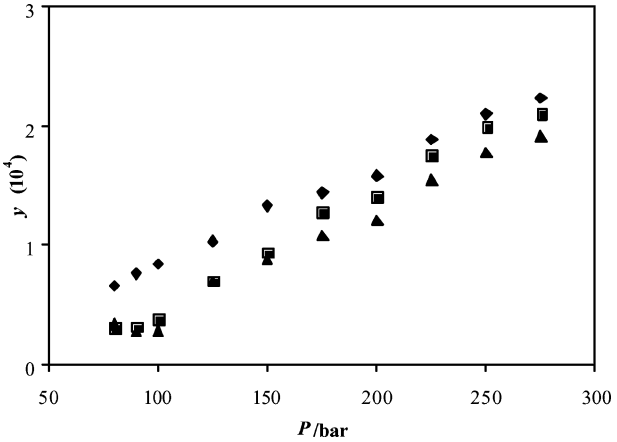

Figure 3. Mole fraction solubility (y) of pindolol in liquid and supercritical carbon dioxide: ◆, 298 K; ■, 308 K; ▲, 318 K.

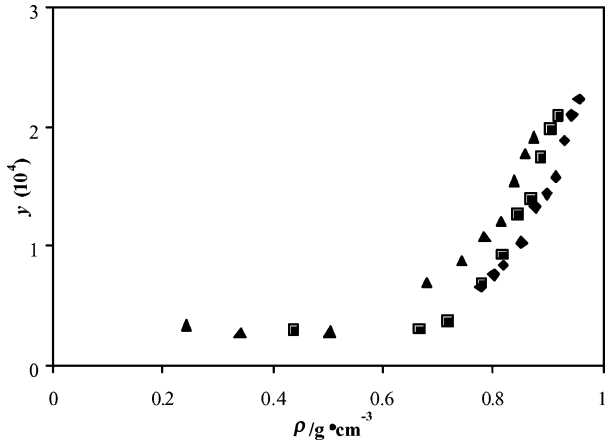

Figure 4. Mole fraction solubility (y) of pindolol in liquid and supercritical carbon dioxide as a function of density (ρ): ◆, 298 K; ■, 308 K; ▲, 318 K.

and placed in the vessel at the low-density measurements was on the same order as the accuracy of the analytical balance; therefore, those data have a large error.

Acebutolol, atenolol, nadolol, and propranolol all formed insoluble salts when exposed to carbon dioxide between (298 and 318) K and pressures between (80 and 275) bar. Francis[11] and Dandge et al.[12] both found that the weakly acidic carbon dioxide can have an affinity for strong basic amines. Ammonia ($K_b = 10^{-5}$), aliphatic amines ($K_b = 10^{-4}$), and pyrrolidine ($K_b = 10^{-9}$) all formed salts when exposed to carbon dioxide. Several weaker bases—aniline ($K_b = 4 \times 10^{-10}$), pyridine ($K_b = 2 \times 10^{-10}$), and 2-picoline ($K_b = 9 \times 10^{-9}$)—dissolved in carbon dioxide and did not salt out. Pyrrolidine with $K_b = 10^{-9}$ did dissolve in carbon dioxide but also formed some insoluble salts;[12] therefore, Dandge et al. postulated that there is an upper limit on the basicity for amine compounds that will allow them to be soluble in carbon dioxide. For the simple amine structures they explored, they believed that a K_b of 10^{-9} or higher is required to have solubility in compressed carbon dioxide. All of the β blockers we explored had at least one secondary amine group that should greatly influence the K_b of the compound. The K_b values of all of the β blockers are presented in Table 2.

Pindolol is less basic than the other compounds; therefore, it would be the least probable to form a salt with carbon dioxide. Its K_b is significantly higher than the cutoff suggested by Dandge et al.[12] for being soluble in carbon dioxide. However, the β blockers we examined are of the

Table 2. K_b (10^5) of β Blockers

acebutolol[17]	2.6
atenolol[17,18]	2.8
nadolol[17]	4.7
pindolol[18]	0.63
propranolol[18]	3.4

3-(aryloxy)-1-(alkyalminio)-2-propanol type, which are much more complex than the simple amines explored by Dandge et al.[12] The presence of an ether group has been shown to increase solubility,[19] and although the indole group on pindolol greatly increases its polarity,[5] it is still able to be dissolved in carbon dioxide. Pindolol's ability to be dissolved is most likely due to its low K_b combined with its favorable interactions between the carbon dioxide and the ether linkage.

Conclusions

The solubility measurements of pindolol in carbon dioxide at (298, 308, and 318) K from (80 to 275) bar have been determined. Solubility increased with increasing density and also increased slightly with increasing temperature. Four β blockers with higher K_b values—acebutolol, atenolol, nadolol, and propranolol—all reacted with carbon dioxide to form yellow-brown salt complexes that were not soluble in carbon dioxide over the conditions tested. Although all of the β blockers tested contain an ether linkage that has been shown to have favorable interactions with carbon dioxide and increase solubility, the basicity of the 3-(aryloxy)-1-(alkyalminio)-2-propanol compounds dominates whether salting will occur. For this type of compound, the K_b should be lower than 10^{-5} to be soluble.

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